REMARKS

In the Office Action, the Examiner objected to claim 11 for informality, rejected claims 13-19 and 25-30 for being indefinite, and rejected claims 1-3, 7-14, 16-20, and 25 for being anticipated by Zhang et al. (2001 Biofactors 15:27-38). The Examiner indicated that claims 4-6, 15, and 24 are allowable if rewritten in independent form.

Each issue raised by the Examiner is considered separately below. In view of the amendments noted above and the remarks below, the applicants respectfully request reconsideration of the merits of this patent application.

Amendment to the Specification

Paragraph [0001] on page 1 of the application has been amended to correct a typographical error in the year in which the priority provisional application is filed.

Claim Objection

The Examiner objected to claim 11 for informality noting that the claim contains an extra comma after the term "rejection reaction." The extra comma has been deleted and the objection is believed to have been overcome.

Indefiniteness Rejections under 35 U.S.C. §112, Second Paragraph

The Examiner rejected claims 13-19 and 25-30 under 35 U.S.C. §112, second paragraph for being indefinite. In particular, the Examiner alleged that the phrase "in an amount sufficient to kill the cell" as recited in claims 13-19 and 25 and the phrase "suitable wave length" as recited in claims 26 and 30 (and thus also claims 27-29) are not clearly defined.

The applicants respectfully submit whether a claim is definite should be viewed from the standpoint of a skilled artisan. Claims 13-17 have been amended to limit the cells to cancer cells and claims 18 and 19 have been canceled. As described in the instant application, the inventors have found that Se(0) preferentially kills cancer cells over normal cells. A skilled artisan understands and appreciates that the amount of Se(0) needed to kill cancer cells can vary depending on the particular type of cancer cell being targeted and the circumstances under which Se(0) is used. For example, solid tumors tend to require higher amounts than leukemia cells and short exposure times typically require higher amounts than

long exposure times. The application also demonstrated that treatment temperature affects the amount of Se(0) needed to kill leukemia cells (see Fig. 9). For a particular type of cancer cell and a particular set of treatment conditions, whether a specific amount of Se(0) is sufficient to kill the cancer cells can be readily determined by a skilled artisan if not already apparent from the disclosure of the present application. Therefore, amended claims 13-17 is definite to a skilled artisan. The same arguments apply to claim 25.

Similarly, a skilled artisan understands and appreciates that the wavelength of a light suitable for oxidizing a selone dye in the presence of oxygen can vary depending on the particular selone dye being used. It is well understood in the art that different selone dyes have different absorption spectra and what is required for the oxidization of a particular selone dye is at least a partial overlap between the emission spectrum of the light source and the absorption spectrum of the dye. It is well within the capability of a skilled artisan to determine the adsorption of a particular selone dye and thus whether a particular wavelength is suitable for oxidizing the dye. Therefore, claims 26-30 are definite to a skilled artisan.

Rejections under 35 U.S.C. §102

The Examiner rejected claims 1-3, 7-14, 16-20, and 25 under 35 U.S.C. §102(b) for being anticipated by *Zhang et al.* (Biofactors 15: 27-38, 2001).

First, the Examiner asserts that claims 1-3 and 7-12 are anticipated because *Zhang et al.* disclose Se(0) particles of 20-60 nm in size (page 28, method 2.1; relevant to claims 1-3 and 8-10) and the use of nano-Se(0) on human heptoma HepG2 cells (page 28, method 2.2; relevant to claims 7, 11, and 12). Claims 1 and 3 have been canceled and claim 2 (and thus claims 7-12) has been amended to depend on claim 4, which limits the size of Se(0) particles to 0.4-5 nm. The rejection against claims 1-3 and 7-12 is believed to have been overcome.

The Examiner next asserts that claims 20-23 are anticipated by *Zhang et al.* because *Zhang et al.* teach that Se(0) can protect cells against paraquat-induced cell death (page 32, results 3.2 and Fig. 5) and further indicate that the protected cells contain glutathione (GSH) (page 32, result 3.4). The applicants note that claims 20-23 of the instant application are directed at using Se(0) to sensitize cells to other cytotoxic agents and as a result promoting the cell killing activity of these agents. This is the opposite to using Se(0) to protect cells from a cytotoxic agent (e.g., paraquat)-induced cell death taught by *Zhang et al.* Therefore, claims 20-23 are not anticipated by *Zhang et al.*

The Examiner next asserts that claim 25 is anticipated by *Zhang et al.* because *Zhang et al.* teach that the first reaction of adsorbed Se *in vivo* is with GSH to form selenodiglutathione (page 32, results 3.4) and Fig. 7 of *Zhang et al.* shows that the ratio of reacted GSH responded linearly to increasing Nano-Se concentrations (page 33). The applicants respectfully submit that the above parts of *Zhang et al.* do not teach the subject matter of claims 25.

Claim 25 of the instant application is directed at using Se(0) to reduce intracellular GSH level. *Zhang et al.* do not provide any original data and only cite *Kuchan and Milner* (reference 20 in *Zhang et al.*) to state that the first reaction of adsorbed Se *in vivo* is with GSH to form selenodiglutathione (at page 32, results 3.4). *Kuchan and Milner* are concerned with selenite, not elemental selenium (Se(0)), and the adsorbed Se referred to in *Kuchan and Milner* is adsorbed selenite, not Se(0) as recited in claim 25. One cannot predict whether Se(0) will first react with GSH *in vivo* based on the indication that selenite does, especially given that the Se in selenite is Se^{4+}, which is a much stronger oxidizing agent than Se(0).

With regard to Fig. 7 of *Zhang et al.* showing that the ratio of reacted GSH responds linearly to increasing Nano-Se concentrations, the applicants believe that Fig. 8 rather than Fig. 7 should have been referred to in this regard and *Zhang et al.* made a typographical error in referring to Fig. 7 (page 32, results 3.4). Fig. 8 shows the result of a cell free system (page 29, method 2.4) and is thus different from the subject matter of claim 25 of reducing intracellular GSH levels. Therefore, claim 25 is not anticipated by *Zhang et al.* GSH is the only reducing agent employed in the cell free system of *Zhang et al.* and the result from such a system cannot be extrapolated to the *in vivo* situation as there are many other molecules in a cell that may interact with Se(0) and one would not have been able to predict whether Se(0) would be titrated out by these other molecules *in vivo* before being able to react with GSH.

Finally, the Examiner asserts that claims 13, 14, and 16-19 are anticipated by *Zhang et al.* because *Zhang et al.* showed that Se(0) killed a number of mice in a toxicity experiment (page 34, Table 1). Claims 13 (and thus claims 14, 16, and 17) has been amended to limit the cells to cancer cells. Claims 18 and 19 have been canceled.

As discussed earlier, the inventors found that Se(0) preferentially kills cancer cells over normal cells and therefore is useful as an anticancer agent for killing cancer cells but not the human or nonhuman animal host. The toxicity experiment of *Zhang et al.* tested the toxicity of Se(0) on "normal" mice (normal mouse cells) and showed that Se(0) killed

"normal" mice (Table 1). Furthermore, in the experiment in which *Zhang et al.* tested the effect of Se(0) on cancer cells, it was shown that even at a very high concentration of 100 μM Se(0) did not kill human heptoma HepG2 cells but only slowed down their growth (Fig. 6). Therefore, amended claims 13, 14, 16, and 17 as well as new claims 48, 49, and 51 are not anticipated by *Zhang et al.* If *Zhang et al.* teach anything, it is that Se(0) may kill a human or animal host first before it kills the tumor cells.

Allowable Subject Matter

The Examiner objected to claims 4-6, 15, and 24 for being dependent upon rejected claims and indicated that they would be allowable if rewritten in independent form. Given that claim 15 was also rejected for being indefinite in the Office Action (see above), it is believed that the Examiner meant that claims 4-6 and 24 would be allowable if rewritten in independent form. In this regard, claims 4-6 and 24 have been rewritten in independent form. In addition, claim 2 (and thus claims 7-12) has been amended to depend on claim 4. Accordingly, claims 2, 4-12, and 24 are believed to be allowable.

The remaining pending claims, i.e. claims 13-17, 20-23, 25-30, and new 48-51 are also believed to be allowable for the reasons provided above.

No extension of time is believed to be necessary and no fee is believed to be due in connection with this response. However, if any extension of time is required in this or any subsequent response, please consider this to be a petition for the appropriate extension and a request to charge the petition fee to Deposit Account No. 17-0055. No other fee is believed to be due in connection with this response. However, if any fee is due in this or any subsequent response, please charge the fee to the same Deposit Account No. 17-0055.

Respectfully submitted,

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